



US Ser. No. 09/673,779  
Currently Pending Claims

-1-

WHAT IS CLAIMED IS:

1. (Amended) A method for identifying the presence of a bacterium in a sample comprising
  - a) testing said sample by Gram-staining and
  - b) testing said sample with a probe according to an *in situ* hybridisation protocol selected on the basis of the outcome of said Gram-staining and identifying the presence of the bacterium in the sample.
2. A method according to claim 1 wherein said sample is a clinical sample.
3. (Amended) A method according to claim 2 wherein said sample is mammalian blood.
4. (Amended) A method according to claim 1 when said Gram-staining indicates the presence of a Gram-negative bacterium in said sample, further comprising determining the rod or coccus character of said bacterium.
5. (Amended) A method according to claim 4 wherein said character is of the rod type, further comprising hybridising said sample with at least one probe selected from a group consisting of probes capable of hybridising with nucleic acid found *Escherichia coli*, in *Klebsiella pneumoniae*, in *Klebsiella oxytoca*, in *Serratia marcescens*, in *Enterobacter aerogenes*, in *Enterobacter cloacae*, in *Proteus vulgaris*, in *Proteus mirabilis*, in *Salmonella typhi*, in *Pseudomonas aeruginosa*.
6. A method according to claim 5 wherein said nucleic acid is ribosomal RNA.
7. (Twice Amended) A method according to claim 6 wherein said probe is having no more than five, preferably no more than two mismatches with a probe selected of a group composed of probes having a sequence  
GCCTGCCAGTTCGAATG (SEQ ID NO:1) or  
GTAGCCCTACTCGTAAGG (SEQ ID NO:2) or
8. A method according to claim 4 wherein said character is of the coccus

type, further comprising subjecting said sample to treatment with a lysis buffer comprising lysozyme.

9. (Amended) A method according to claim 1, when said Gram-staining indicates the presence of a Gram-positive bacterium in said sample, further comprising determining the rod or coccus character of said bacterium.

10. A method according to claim 9 wherein said character is of the rod type, further comprising subjecting said sample to treatment with a lysis buffer comprising lysozyme and/or Proteinase K.

11. (Amended) A method according to claim 9 wherein said character is of the coccus type, further comprising determining a chain-like or clump-like character of said bacteria.

12. A method according to claim 11 wherein said character is chain-like, further comprising subjecting said sample to treatment with a lysis buffer comprising lysozyme.

13. (Amended) A method according to claim 12 further comprising hybridising said sample with at least one probe selected from a group consisting of probes capable of hybridising with nucleic acid found in *Enterococcus faecalis*, in *Streptococcus pneumoniae*, in *Streptococcus mitis*, in *Streptococcus viridans*, in *Streptococcus ganguis*, in *Enterococcus faecium*.

14. A method according to claim 13 wherein said nucleic acid is ribosomal RNA.

15. (Twice Amended) A method according to claim 14 wherein said probe is having no more than five mismatches with a probe selected of a group composed of probes having a sequence TTATCCCCCTCTGATGGG (SEQ ID NO:5) or AGAGAAAGCAAGCTTCTCGTCCG (SEQ ID NO:10) or GCCACTCCTTTTCCGG (SEQ ID NO:7).

16. A method according to claim 11 wherein said character is clump-like, further comprising subjecting said sample to treatment with a lysis buffer

Amended - A method according to claim 11, wherein said character is clump-like.

hybridising said sample with at least one probe selected from a group consisting of probes capable of hybridising with nucleic acid found in *Staphylococcus*

*aureus*, in *Staphylococcus haemolyticus*, in *Staphylococcus saprophyticus*.

18. A method according to claim 17 wherein said nucleic acid is ribosomal RNA.

19. (Twice Amended) A method according to claim 18 wherein said probe is having no more than five mismatches with a probe selected of a group consisting of probes having a sequence GCTAATGCAGCGCGATCC (SEQ ID NO:8) or CCGAAGGGGAAGGCTCTA (SEQ ID NO:9) or AGAGAAGCAAGCTTCTCGTCCGTT (SEQ ID NO:10).

20. (Amended) A method according to claim 4 further comprising hybridising said sample with at least one positive control probe and/or with at least one negative control probe.

21. (Amended) A method according to claim 20 wherein said positive control probe comprising no more than five mismatches with a probe with the sequence GCTGCCTCCCGTAGGAGT (SEQ ID NO:11) and/or wherein said negative control probe comprises no more than five mismatches with a probe with the sequence ACTCCTACGGGAGGCAGC (SEQ ID NO:12).

22. (Amended) A method according claim 1 further comprising a one-step procedure of binding bacteria present in said sample to a microscopic slide and simultaneously fixing intracellular structures.

23. (Amended) A method according claim 1 wherein said probe is selected for its properties of reactivity with a selected one or more of bacterial genera and/or species including a consideration of the susceptibility to antibiotic treatment of said probe.